This report is required by law (7 USC 2143). Failure to report according to the regulations can result in an order to cease and desist and to be subject to penalties as provided for in Section 21

See attached form for additional information.

Interagency Report Control No.:

UNITED STATES DEPARTMENT OF AGRICULTURE
ANIMAL AND PLANT HEALTH INSPECTION SERVICE

ANNUAL REPORT OF RESEARCH FACILITY
(TYPE OR PRINT)

1. CERTIFICATE NUMBER: 55-R-0001
CUSTOMER NUMBER: 825

FORM APPROVED
OMB NO. 0579-0036

Wake Forest University
Medical Center Boulevard
Winston Salem, NC 27109

Telephone: (b)(6), (b)(7)c

3. REPORTING FACILITY (List all locations where animals were housed or used in actual research, testing, or experimentation, or held for these purposes. Attach additional sheets if necessary)

FACILITY LOCATIONS (Sites) - See Atached Listing

REPORT OF ANIMALS USED BY OR UNDER CONTROL OF RESEARCH FACILITY (**Attach additional sheets if necessary or use APHIS Form 7023A**)

A. Animals Covered By The Animal Welfare Regulations	B. Number of animal being bred, conditioned, or held for use in teaching, testing, experiments, research, or surgery but not ye used for such purposes.	C. Number of animals upon which teaching, research, experiments, or tests were conducted involving no pain, distress, or use o pain-relieving drugs.	D. Number of animals upon which experiments, teaching, research, surgery, or tests were conducted involving accompanying pain or distress to the animals an for which appropriate anesthetic, analgesic, or tranquilizing drugs were used.	E. Number of animals upon which teaching, experiments, research, surgery or tests were conducted involving accompanying pain or distress to the animals and for wh the use of appropriate anesthetic, analgesic, or tranquiliz drugs would have adversely affected the procedures, res or interpretation of the teaching, research, experiments, surgery, or tests. (An explanation of the procedures producing pain or distress in these animals and the reas such drugs were not used must be attached to this report	F. TOTAL NUMBER OF ANIMALS (COLUMNS C + D + E)
4. Dogs			12		12
5. Cats			63		63
6. Guinea Pigs			0		0
7. Hamsters			0		0
8. Rabbits			190		190
9. Non-human Primates		207	1315	13	1535
10. Sheep			310		310
11. Pigs			153		153
12. Other Farm Animals			0		0
13. Other Animals					
Bats		17			17
Squirrels		62			62
Ferrets		60	54		114

ASSURANCE STATEMENTS

1) Professionally acceptable standards governing the care, treatment, and use of animals, including appropriate use of anestetic, analgesic, and tranquilizing drugs, prior to, during, and following actual rese teaching, testing, surgery, or experimentation were followed by this research facility.

2) Each principal investigator has considered alternatives to painful procedures.

3) This facility is adhering to the standards and regulations under the Act, and it has required that exceptions to the standards and regulations be specified and explained by the principal investigator and ap Institutional Animal Care and Use Committee (IACUC). **A summary of all such exceptions is attached to this annual report.** In addition to identifying the IACUC-approved exceptions, this summary inc brief explanation of the exceptions, as well as the species and number of animals affected.

4) The attending veterinarian for this research facility has appropriate authority to ensure the provision of adequate veterinary care and to oversee the adequacy of other aspects of animal care and use.

CERTIFICATION BY HEADQUARTERS RESEARCH FACILITY OFFICIAL
(Chief Executive Officer or Legally Responsible Institutional Official)

SIGN (b)(6), (b)(7)c P DATE SIGNED 11/24/08

APHIS

NOV 25 2008

This report is required by law (7 USC 2143). Failure to report according to the regulations can result in an order to cease and desist and to be subject to penalties as provided for in Section 2150

See reverse side for additional information

Interagency Report Control No 0160-DOA-AN

UNITED STATES DEPARTMENT OF AGRICULTURE
ANIMAL AND PLANT HEALTH INSPECTION SERVICE

CONTINUATION SHEET FOR ANNUAL REPORT OF RESEARCH FACILITY

(TYPE OR PRINT)

1. REGISTRATION NO. 55-R-0001 CUSTOMER NO. 825

FORM APPROVED OMB NO 0579-0036

2 HEADQUARTERS RESEARCH FACILITY *(Name and Address as registered with USDA, include Zip Code)*

Wake Forest University
Medical Center Boulevard
Winston-Salem, NC 27109

REPORT OF ANIMALS USED BY OR UNDER CONTROL OF RESEARCH FACILITY *(Attach additional sheets if necessary or use this form.)*

A. Animals Covered By The Animal Welfare Regulations 13. Other Animals	B. Number of animals being bred, conditioned, or held for use in teaching, testing, experiments, research, or surgery but not yet used for such purposes.	C. Number of animals upon which teaching, research, experiments, or tests were conducted involving no pain, distress, or use of pain-relieving drugs	D. Number of animals upon which experiments, teaching, research, surgery, or tests were conducted involving accompanying pain or distress to the animals and for which appropriate anesthetic, analgesic, or tranquilizing drugs were used.	E. Number of animals upon which teaching, experiments, research, surgery or tests were conducted involving accompanying pain or distress to the animals and for which the use of appropriate anesthetic,analgesic, or tranquilizing drugs would have adversely affected the procedures, results, or interpretation of the teaching, research, experiments, surgery, or tests *(An explanation of the procedures producing pain or distress in these animals and the reasons such drugs were not used must be attached to this report)*	F. TOTAL NO OF ANIMALS (Cols. C + D + E)
Chinchillas		380			380

ASSURANCE STATEMENTS

1) Professionally acceptable standards governing the care, treatment, and use of animals, including appropriate use of anesthetic, analgesic, and tranquilizing drugs, prior to, during, and following actual research, teaching, testing, surgery, or experimentation were followed by this research facility.

2) Each principal investigator has considered alternatives to painful procedures.

3) This facility is adhering to the standards and regulations under the Act, and it has required that exceptions to the standards and regulations be specified and explained by the principal investigator and approved by the Institutional Animal Care and Use Committee (IACUC). **A summary of all the exceptions is attached to this annual report.** In addition to identifying the IACUC-approved exceptions, this summary includes a brief explanation of the exceptions, as well as the species and number of animals affected.

4) The attending veterinarian for this research facility has appropriate authority to ensure the provision of adequate veterinary care and to oversee the adequacy of other aspects of animal care and use.

CERTIFICATION BY HEADQUARTERS RESEARCH FACILITY OFFICIAL
(Chief Executive Officer or Legally Responsible Institutional official)
I certify that the above is true, correct, and complete (7 U.S.C. Section 2143)

SIGNATURE OF C.E.O. OR INSTITUTIONAL OFFICIAL	NAME & TITLE OF C.E.O. OR INSTITUTIONAL OFFICIAL *(Type or Print)*	DATE SIGNED

Attachment to APHIS FORM 7023

Explanation of Column E Procedures
Wake Forest University 55-R-0001
Fiscal Year 2007-2008

The following protocols involve experiments reported in Column E. Experiments were approved by the Wake Forest University (WFU) Animal Care and Use Committee (ACUC), after their determination of scientific justification.

1. *A05-136/A08-137: Neurophysiological Recording from Nonhuman Primate Brain*
 Number of animals used in this study: ***13***
 Species (common name) of animals used in the study: *Male rhesus monkeys (Macaca mulatta)*
 Explain the procedure producing pain and/or distress:
 One of the aims of this study is to identify those brain areas most affected by sleep deprivation in an animal model. To that end, monkeys are kept awake for 36-54 hours by use of nonaversive stimuli (lights, videos, music, food treats, toys, random computer sounds, interactions with technicians). Behavioral, electrophysiological, and brain metabolic parameters measured after deprivation are compared to baseline parameters. Sleep deprivation studies are performed at minimum 10 day intervals, typically only once per 2-4 weeks per monkey. While the procedures used to produce sleep deprivation are not themselves considered painful, it is recognized that the lack of sleep may cause monkeys to experience some unrelieved distress during the 36-54 hours when they are kept awake. However, such distress is not entirely unrelieved, since the monkeys are allowed to sleep once the physiological and behavioral assessments are completed. In addition, sleep deprivation procedures utilize nonaversive alerting stimuli, constant interaction with technician staff with whom the monkeys are most familiar, and maintainance of social interaction by working with two or more monkeys at a time. Upon completion of the procedure, the monkeys are returned to their home cage and normal social environment and allowed to sleep. They are also given a normal 12 hour sleep cycle that night.

 Provide scientific justification why pain and/or distress could not be relieved. State methods used to determine that pain and/or distress relief would interfere with test results. (For Federally mandated testing, see next response.):
 Although humans can undergo brief periods with little to no sleep, the problems of extended sleep deprivation are vitally important especially for military personnel, shift-workers, medical personnel, mothers with very young children, and long-distance truck drivers, all of whom must continue to do their essential duties with prolonged periods of sleep loss or interruption. Despite such sleep deprivation, societal and job demands require these individuals to be attentive, remember accurately, and make appropriate decisions. Before an effective means of alleviating the effects of sleep deprivation on memory, attention, and decision-making can be developed, it is necessary to identify those cognitive functions and brain areas most affected by sleep deprivation. This ACUC approved study is necessary to characterize the effects of sleep deprivation in an animal model that is quite similar to that of a human performing a job that requires hand-eye coordination, short-term memory, and decisions based on information presented in the task. The experiment provides access to information (i.e. electrophysiological measures and testing of paliative drugs) that cannot be obtained in human studies. The procedures themselves are not painful, but the actual experience of being sleep-deprived <u>may</u> result in distress to the monkeys, which would be unrelieved during the 36-54 hours that they are kept awake. However, since the stress of sleep deprivation is precisely the effect being studied, it cannot be alleviated during the period of the experiment. Efforts are made to alleviate the distress as soon as the experimental measurements are completed, and sleep deprivation is not repeated for a minimum of 10 days.

 What, if any, federal regulations require this procedure? Cite the agency, the Code of Federal Regulations (CFR) title number and the specific section number (e.g. APHIS, 9 CFR 113.102):
 Agency: CFR:

Attachment to APHIS FORM 7023

Summary of Exceptions to Regulations and Standards
Wake Forest University 55-R-0001
Fiscal Year 2007-2008

1. The following is an exception to the requirements for sanitizing nonhuman primate primary enclosures at required frequencies:

(b)(2)High, (b)(7)f is used to house nonhuman primates imported directly from foreign sources (Indonesia). As required by the Centers for Disease Control (CDC), cages used in this building can not be sent through the mechanical cage washer until after the first thirty-one days (Phase I) of quarantine. Animal waste is removed from cage pans daily. After monkeys have completed Phase I quarantine, cages are sanitized at intervals in accordance with federal regulations and standards.

2. All of the following protocols involve exceptions to the cage size requirements for sheep primary enclosures. Each exception was approved by the Wake Forest University (WFU) Instituitional Animal Care and Use Committee (IACUC), after their determination of scientific justification. For each protocol listed, sheep *(Ovis aries)* are used in studies of pituitary-adrenal hormones, renin, and vasopressin, and are housed in metabolism carts for up to fourteen days. These studies are important to the understanding of factors relating to the health and survival of newborn infants. The Committee's justification for the exception is based on data that show movement of animals in and out of the carts on a daily basis would adversely influence the hormones measured in these experiments.

A06-003: Prenatal and Neonatal Pituitary-Adrenal Function,	Number Used: 12
A03-148/A07-126: Prenatal Glucocorticoid and Postnatal Blood Pressure,	Number Used: 0
A04-155/A07-178: Fetal Endocrine - Cardiovascular Relationships,	Number Used: 0
A05-105: Vulnerability of the Fetal Brain to Hypoxia-Ischemi,	Number Used: 11
A05-077/A08-112: Program Project: Prenatal Events-Postnatal Event,s	Number Used: 58

3. The following protocols involve exceptions to the requirements for watering nonhuman primates. For each protocol listed, full access to water is regulated in a manner to ensure animals are adequately motivated to perform traditional operant conditioning paradigms. Each exception was approved by the WFU IACUC, after their determination of scientific justification.

a. *A06-122: Diencephalic Mechanisms of Visuomotor Integration*
Access to water is regulated such that the first fluid intake of the day will occur during a training or experimental session. A well-trained animal will work to satiety during an experimental session and, in doing so, obtain its minimum daily water requirement. In the event that an animal does not obtain its minimum fluid intake during a session, fluid will be supplemented to the appropriate level when the animal is returned to its home cage. Typically, water regulation is begun on Sunday in preparation for behavioral testing on Monday. Animals participate in experiments on Monday through Friday and receive ad libitum access to water on Friday afternoon through Sunday morning. Throughout the duration of an individual animal's participation in experimental protocols, fluid intake, weight, and a variety of other physiological and behavioral measures are recorded to assure the animal's continued health and well-being. Animals are weighed daily (on the days they are tested). Health and hydration are assessed every day by both research and veterinary personnel. These studies are important to understanding the neural bases of sensory and motor control of the mechanisms of decision making in humans.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 6

b. *A04-050/A07-026: Cortical Implications for Rehabilitation*
Access to water is regulated such that the first fluid intake of the day will occur during a training or experimental session. A well-trained animal will work to satiety during an experimental session and, in doing so, obtain its minimum daily water requirement. In the event that an animal does not obtain its minimum fluid intake during a session, fluid will be supplemented to the appropriate level when the animal is returned to its home cage. Typically, water regulation is begun on Sunday in preparation for behavioral testing on Monday. Animals participate in experiments on Monday through Friday and receive ad libitum access to water on Friday afternoon through Sunday morning. Throughout the duration of an individual animal's participation in experimental protocols, fluid intake, weight, and a variety of other physiological and behavioral measures are recorded to assure the animal's continued health and well-being. Animals are weighed daily (on the days they are tested). Health and hydration are assessed every day by both research and veterinary personnel. Findings from this study will aid in improving rehabilitive therapy for individuals following a stroke.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 4

c. *A07-036: Multisensory integration in primate brain - neocortex and superior colliculus.*
Water regulation only occurs during periods of training and experimentation. Throughout the year animals are given ad lib access to water for variable, but often extended, periods of time (months). Prior to initiation of training, each animal's *ad lib* intake of fluid is measured each day to determine its normal daily fluid consumption; generally this is between 20-60 ml/kg. Then, during training and experiments, the animal is allowed to work to satiety during a session. If the animal will not work for that long, which can occur particularly early in the training, its fluid intake is supplemented with either pieces of fruit (apple, orange) or Gatorade. On the weekends, each animal is given free access to water *ad lib* in its cage. When on study, body weight is recorded at least 3 times per week. A daily record is made of the animal's fluid intake as well as the presence of urine and feces in the pan. Health and hydration are assessed every day by both research and veterinary personnel. These studies will provide information about the dynamics of brain function in multisensory integration from external stimuli, e.g., light and sound and eye movements.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 3

d. *A07-037: Subcortical neural basis of auditory behavior.*
Water intake is regulated so that the animals' first fluid intake of the day occurs during a training or experimental session. Fully-trained animals work to satiety, obtaining more than their minimum fluid requirement. In the event that an animal does not obtain its minimum intake during a session (particularly during the early stages of training) it is supplemented after the end of the session by the investigator. Animals participate in experiments 5 days a week and have free access to water for two days per week. Prior to beginning training, each animal's ad lib intake of fluid is recorded to determine its normal daily consumption. This is generally in the range of 20-60 ml/kg. A daily record is made of the animal's fluid and food intake. Body weights are recorded at least twice per week (daily during experiments). Health and hydration are assessed every day by both research and veterinary personnel. Water regulation is temporarily suspended if a monkey exhibits signs of dehydration (i.e. decreased urine production, dry feces, anorexia, changes in skin elasticity). Findings from this project will offer insight into possible mecahnisms that the brain utilizes to hear sounds in complex (noisy) environments, and will offer insights that allow us to design better assistive hearing devices such as hearing aids and cochlear implants.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 2

e. *A06-033: Neurophysiological effects of training in visual cognitive tasks*

Water intake is regulated so that the animals' first fluid intake of the day occurs during a training or experimental session. Fully-trained animals work to satiety, obtaining more than their minimum fluid requirement. In the event that an animal does not obtain its minimum intake during a session (particularly during the early stages of training) it is supplemented after the end of the session by the investigator. Animals participate in experiments 5 days a week and have free access to water for two days per week. Prior to beginning training, each animal's ad lib intake of fluid is recorded to determine its normal daily consumption. This is generally in the range of 20-40 ml/kg. A daily record is made of the animal's fluid and food intake. Body weights are recorded at least twice per week (daily during experiments). Health and hydration are assessed every day by both research and veterinary personnel. Water regulation is temporarily suspended if a monkey exhibits signs of dehydration (i.e. decreased urine production, dry feces, anorexia, changes in skin elasticity). These studies will provide information about the biological basis of memory in healthy subjects, and suggest the means to modify and compensate for loss of memory function in stroke and brain trauma.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 3

f. *A06-034: The role of the prefrontal cortex in memory and higher cognitive functions*
Water intake is regulated so that the animals' first fluid intake of the day occurs during a training or experimental session. Fully-trained animals work to satiety, obtaining more than their minimum fluid requirement. In the event that an animal does not obtain its minimum intake during a session (particularly during the early stages of training) it is supplemented after the end of the session by the investigator. Animals participate in experiments 5 days a week and have free access to water for two days per week. Prior to beginning training, each animal's ad lib intake of fluid is recorded to determine its normal daily consumption. This is generally in the range of 20-40 ml/kg. A daily record is made of the animal's fluid and food intake. Body weights are recorded at least twice per week (daily during experiments). Health and hydration are assessed every day by both research and veterinary personnel. Water regulation is temporarily suspended if a monkey exhibits signs of dehydration (i.e. decreased urine production, dry feces, anorexia, changes in skin elasticity). Findings from this project will offer insight into possible means of reversing the effects in humans of brain damage and mental illness.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 2

g. A07-127 *Cholinergic influences on prefrontal cortical activity during cognitive function*
Water intake is regulated so that the animals' first fluid intake of the day occurs during a training or experimental session. Fully-trained animals work to satiety, obtaining more than their minimum fluid requirement. In the event that an animal does not obtain its minimum intake during a session (particularly during the early stages of training) it is supplemented after the end of the session by the investigator. Animals participate in experiments 5 days a week and have free access to water for two days per week. Prior to beginning training, each animal's ad lib intake of fluid is recorded to determine its normal daily consumption. This is generally in the range of 20-40 ml/kg. A daily record is made of the animal's fluid and food intake. Body weights are recorded at least twice per week (daily during experiments). Health and hydration are assessed every day by both research and veterinary personnel. Water regulation is temporarily suspended if a monkey exhibits signs of dehydration (i.e. decreased urine production, dry feces, anorexia, changes in skin elasticity). These studies will uncover the results of low levels of acetylcholine (which occur in Alzheimer's disease) and how these effects can be reversed by drugs that have the opposite effect. The study will also provide a novel model for the evaluation of other types of drugs, as they relate to cognitive performance and rehabilitation.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 2

4. The following protocol involves an exception to the lighting and watering requirements for nonhuman primates. The exceptions were approved by the WFU ACUC, after determination of scientific justification.

A05-136/A08-137: Neurophysiological Recording from Nonhuman Primate Brain
Full access to water is regulated in a manner to ensure animals are adequately motivated to perform traditional operant conditioning paradigms. Access to water is regulated such that the first fluid intake of the day will occur during a training or experimental session. A well-trained animal will work to satiety during an experimental session and, in doing so, obtain its minimum daily water requirement (as determined by a formula based on age, size and weight of the animal). In the event that the animal does not obtain its minimum fluid intake during a session, fluid is supplemented to the appropriate level when the animal is returned to its home cage. Typically, water regulation is begun on Sunday afternoon in preparation for behavioral testing on Monday. Animals participate in experiments on Monday through Friday and may receive additional water (or high-water-content fruit and vegetable supplements) on Friday afternoon through Sunday afternoon. Throughout the duration of an individual animal's participation in experimental protocols, fluid intake, body weight, behavioral performance, and observations of animal health are recorded daily. Periodic veterinary assessment, and other measures such as urine output, hematocrit, etc. are also used to assure the animals' continued health and well-being. Animals are weighed on the days they are tested (or at least weekly). Health and hydration are assessed every day by both research and veterinary personnel.

One of the aims of this study is to identify those brain areas most affected by brief (36-54 hour) sleep deprivation in an animal model. A monkey performs its behavioral/recording session normally during the day before sleep deprivation. That evening, before the lights go off in the housing area, the monkey is moved to a separate room in which the lights stay on all night and is kept awake by use of nonaversive stimuli (videos, music, food treats, toys, random computer sounds, interactions with technicians). The behavioral and electrophysiological testing of the monkey is then repeated the following afternoon. Sleep deprivation studies are performed at minimum 10 day intervals, typically only once per 2-4 weeks per monkey.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 13

5. The following protocols involve exceptions to the lighting and space requirements for cats (male and female domestic shorthair cats *(Felis domesticus).* The exceptions were approved by the WFU ACUC, after determination of scientific justification.

These studies examine the role of visual experience on the development of multisensory processes in the midbrain. From birth, kittens are raised to adulthood in total darkness. Total darkness is required to eliminate all visual experience during development in a reversible manner. Procedures such as enucleation or eyelid-suturing are inadequate either because they are not reversible or allow a significant amount of light to penetrate the lids. A portion of the population is returned to full-spectrum lighting at six to twelve months of age. When in darkness, animals are cared for in accordance with all other standards and regulations. Infrared goggles are used by animal care, veterinary, and research personnel who enter the room daily to perform routine husbandry, health evaluations, and research manipulations. Criteria for monitoring the health and well-being of animals were developed, reviewed, and approved by the IACUC.

Some animals in the room (3 during this reporting period) spend six hours per day together (for about 6 months) in an experimental pen which provides adequate space, but less than the minimum square footage per cat required by the USDA for standard housing. The size of the pen is a critical element in restricting the sensory experience of these animals, since it serves to constrain the minimum and maximum angular disparities of visual and auditory experience. Enlarging the size of the experimental pen would increase

the maximal disparity beyond that which is believed to have an effect on reorganization in the nervous system. It is imperative that experimental animals are placed in the pen together to receive the same environmental manipulation. Any difference in the sensory experiences of the animals would constitute an unresolvable confound in the interpretation of the subsequently gathered data. It is important to note that animals are only in the run for six hours per day, after which they are returned to pens that provide the minimum square footage per cat required by the USDA. Results from this work are important for the improved treatment of children with visual disorders such as blindness, amblyopia, and strabismus.

A06-079: Development of Multisensory Integration,	Number Used: 2
A07-117: Development of Multisensory Integration – Cats	Number Used: 2
A07-189: Experiential Influences on Cross-Modal Developmen	Number Used: 9

6. The following protocol involves an exception to the ambient temperature requirements for nonhuman primates. The exception was approved by the WFU IACUC, after determination of scientific justification.

A05-006/A06-093. Thermoregulation in MDMA Abuse
MDMA or "ecstasy" is a widely known recreational drug of abuse and is commonly abused in warm, crowded environments such as "rave" parties and dance clubs. A major consequence of MDMA use involves elevation of body temperature, which increases the risk of toxicity. Currently there is a critical lack of understanding of the neuronal processes involved in MDMA abuse and subsequent hyperthermia. This study proposes to study MDMA and the neurohormonal systems in nonhuman primate models. Understanding the mechanism involved in MDMA-induced hyperthermia and the underlying neurochemistry will allow for increased public awareness of the potential toxicities associated with MDMA use and provide new means for treating MDMA-mediated thermogenesis and abuse.

Animals are housed in accordance with USDA regulations and standards. Following instrumentation and acclimation to primate chairs, behavioral experiments are carried out in monkeys in ventilated, sound attenuating chambers designed to accommodate primate chairs. During the experimental sessions, the ambient temperature in the chamber is adjusted as needed to maintain the chamber at temperatures up to 31±1°C. Body temperature was recorded immediately after the experimental session. During these experiments, no monkey had a core temperature above 41°C that required intervention or termination of the experiment.

Species Used: Male rhesus monkeys (*Macaca mulatta*) Number Used: 5